

4-DIMETHYLAMINOAZOBENZENE

Measurements Research Branch

Analytical Method

Analyte:	4-Dimethylaminoazobenzene	Method No:	P&CAM 284
Matrix:	Air	Range:	4-2000 $\mu\text{g}/\text{m}^3$ in a 50-liter sample
Procedure:	Collection on glass fiber filter and Gas-Chrom P, elution with 2-propanol, analysis by gas chromatography	Precision:	7%
Date Issued:	8/1/78	Classification:	E (Proposed)
Date Revised:			

1. Synopsis

- 1.1 A known volume of air is drawn through a two-stage sampler consisting of a high-efficiency glass fiber filter followed by a bed of Gas-Chrom P to collect the 4-dimethylaminoazobenzene aerosol and/or vapor.
- 1.2 The filter and sorbent sections of the sampler are transferred to stoppered tubes and the 4-dimethylaminoazobenzene is desorbed with 2-propanol.
- 1.3 An aliquot of this solution is injected into a gas chromatograph.
- 1.4 Peak areas are electronically determined and compared with a calibration curve obtained from injections of standard solutions of 4-dimethylaminoazobenzene.

2. Working Range, Sensitivity and Detection Limit

- 2.1 The analytical range is 0.2-100 $\mu\text{g}/\text{sample}$ (4-2000 $\mu\text{g}/\text{m}^3$ for a 50L air sample) when using 0.5 mL of desorbing solution and 1- μL injections into the gas chromatograph.
- 2.2 In experiments in which atmospheres containing 400 $\mu\text{g}/\text{m}^3$ of 4-dimethylaminoazobenzene in air of 80% relative humidity at 30°C were sampled, less than 10% of the compound was found in the sorbent bed after six hours (65 L). Tests to determine breakthrough of the

sorbent bed during the sampling periods showed no detectable 4-dimethylaminoazobenzene downstream of the sorbent bed. The compound existed primarily as an aerosol.

2.3 The limit of detection is 0.05 µg/sample.

3. Interferences

- 3.1 In solutions containing 4-dimethylaminoazobenzene at 3 ng/µL, the following compounds were found not to interfere when their concentrations were 3 ng/µL: azobenzene, N-methylaniline, N,N-dimethylaniline, 4-nitrobiphenyl, 4-aminobiphenyl, 4-hydroxyazobenzene and the 2'-, 3'- and 4'-methyl derivatives of 4-dimethylaminoazobenzene. 4-Aminoazobenzene has the same retention time as the analyte and does interfere.
- 3.2 Any compound sampled with 4-dimethylaminoazobenzene and having the same retention time as the analyte will interfere with the analysis. This type of interference often can be eliminated by changing the operating conditions of the chromatograph, e.g., by temperature programming or by changing the stationary phase.

4. Precision and Accuracy

- 4.1 The precision of the overall method is 7% relative standard deviation for samples collected at 30°C and 80% relative humidity. This value is based on 25 samples involving volumes of 10 to 70 liters and a sampling rate of 0.2 L/min. Concentrations sampled ranged from 67 to 445 µg/m³.
- 4.2 Filters spiked with 0.2 µg 4-dimethylaminoazobenzene and stored at ~24°C yielded recoveries >82% when desorbed within 28 days. Filters spiked with 3.9 µg yielded recoveries of 100% during that period.
- 4.3 If more than 10% of the total recovered weight is found on the Gas-Chrom P section, sample loss may be indicated. Under these circumstances, the actual ambient concentration may be greater than the sample analysis indicates.
- 4.4 At a flow rate of 0.2 L/min, the linear velocity through the 4-mm inlet of the holder is 26.5 cm/s. It is not known if this throat velocity is sufficient for the capture of all particles.
- 4.5 The accuracy of the method has not been determined.

5. Advantages and Disadvantages

- 5.1 The sampler is a small portable device containing no liquids. This is an advantage for sampling air in a worker's breathing zone without interfering with normal work activities.

5.2 This method has not been field tested.

6. Apparatus

- 6.1 Personal air sampling pump that can be maintained for four hours at a flow rate of 0.2 L/min with the sampler in line. Each pump should be calibrated with a representative sampler in line to minimize errors in volume measurements. A bubble flow meter or other suitable flow measuring device may be used.
- 6.2 Sampler, equivalent to that shown in Figure 1. The sampler consists of two sections: a high-efficiency glass fiber filter, followed by a 50-mg bed of Gas-Chrom P. The first section, a 13-mm glass fiber filter (Spectrograde or Type A-E, Gelman Instrument Co., Ann Arbor, MI, or equivalent) is contained in a 13-mm filter holder (SX00-013-00 Millipore Corp., Bedford, MA, or equivalent). The sorbent tube is a 50-mm section of Pyrex glass, 6.4-mm o.d. x 4-mm i.d. flared on one end, containing the section of Gas-Chrom P (Grade 407, 30/60 mesh, 1 m²/g, 0.26 g/cc; Applied Science Laboratories, State College, PA, or equivalent). The sorbent is held by 3.5-mm diameter, 100 mesh, stainless steel screens and 4-mm o.d. Teflon rings, as shown in Figure 1. To connect the two stages, the filter holder is pressed into the flared end of the sorbent tube. The pressure drop across the sampler at 0.2 L/min is ~20 torr. Flexible vinyl caps are used to seal the ends of the sampler.
- 6.3 Gas chromatograph equipped with a flame ionization detector (FID) and injection port.
- 6.4 Glass column, 1.8-m x 2-mm i.d. x 6.4-mm o.d., packed with 3% OV-225 on 80/100 mesh Supelcoport, (Supelco Inc., Bellefonte, PA, or equivalent,)
- 6.5 Potentiometric strip chart recorder.
- 6.6 Test tubes, 1-mL, fitted with polyethylene stoppers.
- 6.7 Glass syringes, 5- and 10- μ L.
- 6.8 Pipettes of convenient volumes for the preparation of standard solutions.
- 6.9 Volumetric flasks of convenient volumes for the preparation of standard solutions.
- 6.10 Clinical centrifuge.
- 6.11 Analytical balance.
- 6.12 Test tube shaker, vortex type.

7. Reagents

Whenever possible, reagents used should be ACS reagent grade or better.

7.1 2-Propanol, UV grade, distilled in glass.

7.2 Acetone, distilled in glass.

7.3 Methyl alcohol.

7.4 Chloroform.

7.5 4-Dimethylaminoazobenzene.

8. Procedure

8.1 Cleaning of Equipment. All glassware used for the laboratory analysis is washed with detergent and rinsed with tap water, distilled water, and methyl alcohol.

8.2 Collection and Shipping of Samples

8.2.1 Immediately before sampling, the vinyl caps are removed from the ends of the sampler and saved for resealing after sampling.

8.2.2 The sorbent end of the sampler is connected to the pump with plastic or rubber tubing. The sampler is positioned vertically during sampling. Sampled air must not pass through any tubing before entering the sampler.

8.2.3 The atmosphere is sampled at a flow rate of 0.2 L/min for no longer than six hours. The flow rate and sampling time, or the volume of sampled air, must be measured as accurately as possible.

8.2.4 The temperature, pressure, and if possible, humidity of the atmosphere being sampled are measured and recorded, along with other pertinent data such as location, date, and time of sampling. Condition of high humidity should be noted.

8.2.5 Blank samples are obtained by handling random collection devices in the same manner as in sampling, except that no air is drawn through them.

8.2.6 Samples shipped to the laboratory should be packed tightly to minimize breakage.

8.2.7 If samples of bulk material associated with the process under investigation are to be shipped to the laboratory, they should not be placed in the same container as the air samplers or blanks.

8.3 Analysis of Samples

8.3.1 Place the glass fiber filter in a 1-mL tube. Place the 50-mg section of Gas-Chrom P in a separate test tube. Teflon rings and stainless steel screens associated with the sampler may be analyzed with the Gas-Chrom P stage. Sampler caps may be reused.

8.3.2 Add 0.5 mL of 2-propanol to each vial and cap it.

8.3.3 Allow the vials to stand for one hour, shaking them intermittently in a test tube vibrator. Centrifuge for 10 minutes.

8.3.4 Gas Chromatographic Conditions

Column:	3% OV-225 on 80/100 mesh Supelcoport in 1.8-m x 2-mm i.d. x 6.4-mm o.d. glass tubing.
Column Temperature:	218°C
Injector Temperature:	228°C
Detector Temperature:	228°C
Helium:	24 mL/min
Hydrogen:	85 mL/min
Air:	378 mL/min
Detector:	FID; Attenuation 1 x 2 at low concentrations
Injection Volume:	1.0µL
Efficiency:	3200 theoretical plates
Capacity Ratio:	18.1

8.3.5 Injection. Flush the 5 µL syringe first with acetone (to insure its cleanliness and to wet the barrel and plunger) and then with the sample solution. Carefully (to minimize the intake of particulate matter) draw 1 µL of sample solution into the syringe and inject the aliquot into the gas chromatograph.

8.3.6 Measure the peak area and read the amount of analyte from the calibration curve.

8.4 Efficiency of Desorption

- 8.4.1 Importance of Determination. The efficiency of desorption for 4-dimethylaminoazobenzene can vary from one laboratory to another and also from one batch of Gas-Chrom P or filters to another. Thus, it is necessary to determine the fraction of 4-dimethylaminoazobenzene that can be recovered from these matrices. Since the efficiency of desorption may also vary with the amount of 4-dimethylaminoazobenzene present, determinations should be made for at least two levels in the range of anticipated sample sizes.
- 8.4.2 Procedure for Determining the Efficiency of Desorption. Place the filters and Gas-Chrom P from unused samplers in 1-mL test tubes. Add to each a known amount of 4-dimethylaminoazobenzene in 10 μ L of chloroform using the same microliter syringe for each. Five filters and Gas-Chrom P samples are thus prepared at each of two different levels in the range of interest. Cap the tubes. Treat a parallel blank in the same manner (no analyte is added). Prepare at least three standards at each of the various levels by adding 10 μ L (same syringe as above) of the 4-dimethylaminoazobenzene solutions to 0.5 mL of desorbing solution. Allow them to stand overnight. Analyze the samples and standards as described in Section 8.3. The efficiency of desorption (D) is the average gas chromatographic peak area given by the samples, corrected for the blank if necessary, divided by the average area given by the standards.

9. Calibration and Standardization

- 9.1 For accuracy in preparation of standards, it is recommended that one standard be prepared in a relatively large volume and at a high concentration. The initial standard is prepared by weighing a selected amount of 4-dimethylaminoazobenzene (e.g., 125 mg) into a 250-mL volumetric flask and adding the desorbing solution to the calibration mark. Dilutions encompassing the concentration range of interest down to 0.4ng/ μ L are then made from the stock solution. Solutions should be stored in a refrigerator when not in use.

CAUTION: 4-Dimethylaminoazobenzene Has Been Identified As A Carcinogen. Appropriate Precautions Must Be Taken In handling The Compound To Avoid Personnel Exposure And Area Contamination.

- 9.2 The standard solutions should be analyzed under the same gas chromatographic conditions and during the same time period as the samples. This will minimize inaccuracy due to day-to-day variations in the setting of instrumental conditions.
- 9.3 Calibration curves for 4-dimethylaminoazobenzene are prepared by plotting the peak areas for the standards against their concentration in μ g/0.5mL.

10. Calculations

- 10.1 In the event that the blank produces a peak with the same retention time as the analyte, the analyst should determine the source of the interference and eliminate or compensate for it.
- 10.2 Read the weight (W_1) in μg of 4-dimethylaminoazobenzene present in the sample from the calibration curve.
- 10.3 The corrected weight (W_2) is determined by

$$W_2 = \frac{W_1}{D}$$

where D is the efficiency of desorption for that sampler stage.

- 10.4 Add the weights found in the sampler stages to determine the total weight (W_t) collected.
- 10.5 The concentration of 4-dimethylaminoazobenzene in air (C , $\mu\text{g}/\text{m}^3$) is given by

$$C = \frac{W_t}{V} \times 10^3$$

where V is the volume of air sampled in liters and 10^3 is the factor to convert from liters to m^3 .

11. Reference

Raul Morales, J.F. Stampfer, Jr., R. E. Hermes, E.E. Campbell, and H.J. Ettinger, "Development of Sampling and Analytical Methods for Carcinogens, October 1, 1976 - December 31, 1977," Progress Report LA-7375-PR, National Technical Information Service, Springfield, Virginia, 1978.

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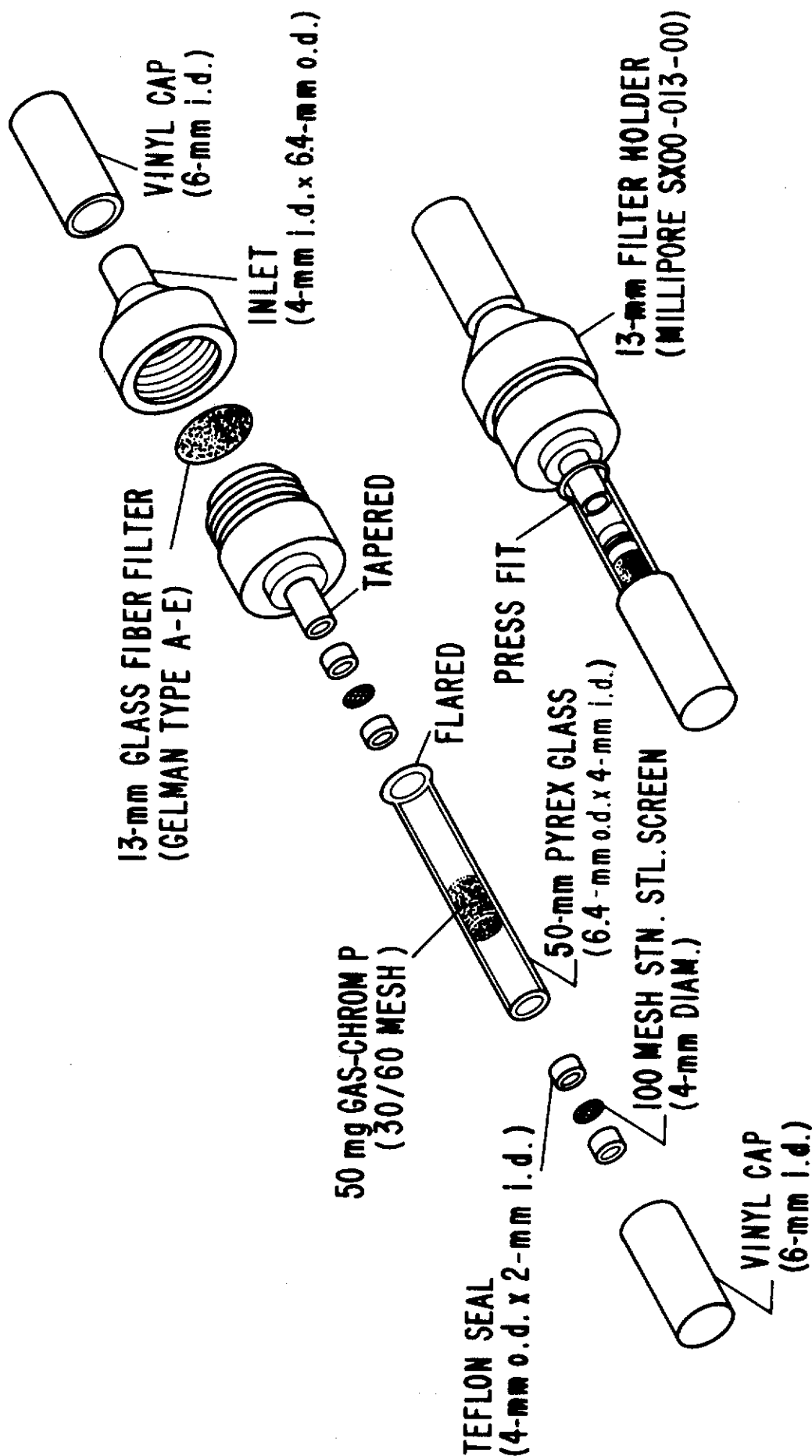


Figure 1. Two-Stage Personal Sampler for 4-Dimethylaminobenzene